

**IN THE SPECIFICATION:**

**Please replace the paragraph beginning at page 4, line 27, and bridging to page 5, line 20, with the following rewritten paragraph:**

The present invention has been made to attain the above object, and includes the following aspects. (1) A method for immobilizing a protein to a solid-phase, which comprises contacting the protein with the solid-phase having hydrophobic surface in the presence of a lower alcohol, a halogenocarboxylic acid and/or a long chain alkyl sulfate. (2) A method for quantitative determination of a protein, which comprises contacting the protein-staining solution with the solid-phase, immobilized with the protein by the method of the above (1), and the determining based on a degree of color development generated thereby. (3) A method for immunoblotting, comprising using the solid-phase to which the protein immobilized by the method of the above (1). (4) A method for detecting an abnormal PrP, which comprises immobilizing an abnormal PrP to a solid-phase by treating a sample to be tested containing the abnormal PrP by the method of the above (1); subsequently reacting with an antibody capable of binding to the abnormal PrP; measuring an amount of an antigen – antibody complex generated thereby, and detecting a presence of an abnormal PrP based on the results thereof. (5) A method for determining prion disease, which comprises detecting an abnormal PrP by the method of the above (4); and determining prion disease based on the results thereof. (6) An reagent solution for immobilizing a protein comprising a low alcohol, a

halogenocarboxylic acid and/or a long chain alkyl sulfate. (7) A kit for detecting an abnormal PrP, which comprises as a constituent reagent: (i) an immobilizing reagent solution 1 containing a lower alcohol, and a halogenocarboxylic acid and/or a long chain alkyl sulfate; (ii) an immobilizing reagent solution 2 containing a lower alcohol, ~~a halogenocarboxylic acid and a nonionic surfactant~~; and (iii) a labeled antibody capable to specifically bind to an abnormal PrP.

**Please replace the paragraph beginning at page 35, line 29, and bridging to page 36, line 8, with the following rewritten paragraph:**

From the viewpoint of ~~[[CV]]~~ Coefficient Variation value (~~CV value~~ = standard variation / mean value (%)) hereinafter designated as CV value) showing the variation of data, it is understood that when the concentration of SDS in the immobilizing reagent solution is lower than 0.1% (W/V), every protein shows the large CV value, that is, the signal intensity is not maintained stably. Contrary, it is indicated that when the SDS concentration in the immobilizing reagent solution is 0.1% (W/V) or more, CV value is relatively small, that is, a value of the signal intensity is maintained stably within such a range of concentration as described above. The results are considered to indicate an amount of a protein immobilized to a solid-phase membrane. Further, although data are not shown, this is confirmed to be reproducible.

**Please replace the paragraph beginning at page 39, line 11, with the following rewritten paragraph:**

The following immobilizing reagent solutions were prepared by using purified water.

Immobilizing reagent solution 1: 0.2% (W/V) SDS, 2.5% (W/V) TCA;

Immobilizing reagent solution 2: 0.2% (W/V) SDS, 2.5% (W/V) TCA, 45% (V/V) ethanol;

Immobilizing reagent solution 3: 0.2% (W/V) SDS, 2.5% (W/V) TCA, 45% methanol (V/V) (Wako Pure Chemical Industries, Ltd., Reagent grade).

**Please replace the paragraph beginning at page 39, line 18, with the following rewritten paragraph:**

Each protein sample in an amount of 20  $\mu\text{L}$  (protein 5  $\mu\text{g}$ ) and 300 ~~[[mL]]~~  $\mu\text{L}$  of a given immobilizing reagent solution were mixed to prepare the immobilization sample 1, the immobilization sample 2 and the immobilization sample 3. The final concentration of each reagent in the immobilization sample is SDS 0.19% (W/V), TCA 2.34% (W/V), ethanol 42.2% (V/V), and methanol 42.2% (V/V).

**Please replace the paragraph beginning at page 40, line 27, and bridging to page 41, line 4, with the following rewritten paragraph:**

Each protein sample in an amount of 20  $\mu\text{L}$  (protein 5  $\mu\text{g}$ ) and 300 ~~[[mL]]~~  $\mu\text{L}$  of a given immobilizing reagent solution were mixed to prepare the immobilization sample 1, the immobilization sample 2 and the immobilization sample 3. The final concentration of each reagent in the immobilization sample is SDS 0.19% (W/V), TCA 2.34% (W/V), TFA 2.34% (W/V), and ethanol 42.2% (V/V).

**Please replace the paragraph beginning at page 41, line 27, and bridging to page 42, line 2, with the following rewritten paragraph:**

Each reagent was dissolved in purified water so that a concentration thereof became 0.2% ~~(V/V)~~ (W/V) SDS, 2.5% ~~(V/V)~~ (W/V) TCA and 45% (V/V) ethanol to prepare the immobilizing reagent solutions.

**Please replace the paragraph beginning at page 48, line 19, with the following rewritten paragraph:**

A final concentration of each reagent in the immobilization sample is SDS 0.094% (W/V), TCA ~~23.4%~~ 2.34 % (W/V) and ethanol 42.2% (V/V), when the protein sample without containing

SDS is used. With regard to the final concentration of each reagent in the case using a protein sample containing SDS, the final concentrations of TCA and ethanol are the same as in the case using the protein sample without containing SDS. Final concentrations of SDS are: 0.16% (W/V) in the case using a protein sample containing 1% SDS; 0.21% (W/V) in the case using a protein sample containing 2% SDS; and 0.34% (W/V) in the case using a protein sample containing 4% SDS.

**Please replace the paragraph beginning at page 49, line 8, with the following rewritten paragraph:**

A final concentration of each reagent in the immobilization sample is SDS 0.19% (W/V), TCA ~~23.4%~~ 2.34% (W/V) and ethanol 42.2% (V/V), when the protein sample without containing SDS is used. Further, a final concentration of each reagent in the case using the protein sample containing 2% SDS is SDS 0.31% (W/V), TCA 23.4% (W/V) and ethanol 42.2% (V/V).

**Please replace the paragraph beginning at page 57, line 1, with the following rewritten paragraph:**

A small tissue pieces of the bovine cerebellum infected with BSE (repeated freeze-thaw), which was the same as that used in Example ~~[[1]]~~ 11 in an amount of 350 mg (wet weight), was homogenized in 1.4 mL of the homogenizing solution (glucose 50 mg/mL) using a multi-beads shocker.

**Please replace the paragraph beginning at page 71, line 13, with the following rewritten paragraph:**

Further, the cutoff value for determination of BSE infection was defined. In the case of Example 13, a value of ~~[[0.185]]~~ 0.175, which was obtained by adding a mean value of the absorbance for the negative specimen performed by the method in Example 13 obtained in Table 9 above (0.035) to a mean value of absorbance for the negative specimen obtained in Example 12, i.e.  $(0.052) + 5SD(5 \times 0.017) =$  ~~[[0.15]]~~ 0.14, was tentatively set as a cutoff value for determining BSE. When the value is applied to the results of Table 8-1 and Table 8-2, a mean value of all absorbance values in Example 13 is higher than the cutoff value, consequently all specimens could be determined as positive.

**Please replace the paragraph beginning at page 75, line 8, with the following rewritten paragraph:**

Each ~~[[600]]~~ 400  $\mu\text{L}$  of the sample for immobilizing PrP prepared above was applied to two wells (duplicate for each sample, i.e. 200  $\mu\text{L}$ / each well) of a multiplate to which a PVDF membrane was set.